

STANFORD UNIVERSITY  
STANFORD, CALIFORNIA

DEPARTMENT OF MEDICAL MICROBIOLOGY

November 11, 1958

Dr. Joshua Lederberg  
Genetics Building  
University of Wisconsin  
Madison 6, Wisconsin

Dear Joshua:

I am amazed that, at a time like this when excitement must be rampant without and within, you can take time to write a thoughtful letter about the hypersensitivity questions in which you are interested. Your questions are good ones, and I will try to do justice to them.

I sent you the manuscript because I believe that it may pertain to the question of earliest cell response to immunologic stimuli which interests you. We made a special report of this work, not because the findings are original in the sense of a new discovery, but in order to try to put into perspective what these reactivities consist of. Uhr and Pappenheimer have been speaking of the induction of delayed hypersensitivity by means of antigen-antibody complexes, as if such complexes in the region of antibody excess have some special property in this respect. Our work was simply to demonstrate, as has been done less pointedly by others in the past, that the reactivity which they see is one which is occasioned by an antigenic stimulus alone; if this is small, the evanescent delayed-appearing response appears to be the only one which the animal is capable of. If it is larger, this is succeeded by humoral antibodies and Arthus (immediate) reactivity.

You ask whether this work is related to that of Tremaine. Her work suggests that animals which have been immunized with protein in a manner calculated to produce circulating antibodies and immediate hypersensitivity can transfer, by means of peritoneal exudate cells, delayed reactivity to normal recipient rabbits. If this is true, it would mean that all animals immunized with ordinary protein antigens actually have a degree of delayed hypersensitivity at all times, and that this does not show up

perhaps when the immunized animal itself is skin-tested later in its course because the larger immediate reactions obscure the delayed ones. Early in the game, before antibodies and immediate reactivity have appeared, the delayed reactions can be seen. If all this is true, it would mean that what we call the Jones-Mote type of early appearing evanescent delayed hypersensitivity actually runs throughout the period of immunologic response and in the sense of persistence would probably be the same as conventional delayed hypersensitivity. Whether this is true or not, for purposes of your interest in the matter it still remains rather apparent that the delayed hypersensitive kind of reactivity seems to be the first response of the animal to an antigen. But to take the matter further, I am not convinced as yet that the "Jones-Mote" delayed hypersensitivity runs throughout the period of immunologic response as Tremaine's work suggests. If this were true, one should expect to be able to show corneal reactions (which everyone agrees occur only when delayed hypersensitivity is present) in any animal which has been immunized with any antigen, and this is not so, as evidenced by the fact that the corneal test is a very good one used to distinguish immediate from delayed hypersensitive states. In our own work with guinea pigs highly anaphylactic to tuberculo-protein (when lipopolysaccharide was not used with it) no corneal responses were seen at all, and this has been general experience. My point is that if she can transfer cells to a recipient animal intravenously and by this means make the cornea of the recipient animal reactive to antigen, why can't the donor of the cells itself show the same kind of reactivity? There is other evidence, but I don't want to prolong this letter to the length of a manuscript.

You ask also how all this--Tremaine's and our own work--may be related to the observations of Rosenberg et al. in a recent issue of the Journal of Immunology. In that case cells of the spleen were transferred to skin sites and later injections of antigen were made into the same sites for the purpose of demonstrating the appearance of small amounts of antibodies manufactured by the transplanted cells. The essential conditions here are the same

as those used by Tremaine, and this confuses the issue. I should say, however, that Rosenberg was observing true immediate reactions for the simple reason that they occurred immediately. The animals were killed fifteen minutes after antigen injection in order to determine the extent of infiltration of the injected site by dye. If Tremaine's work holds up and applies to the skin as well, which it should, I would say that if Rosenberg were to wait for an appropriate period of hours after the injection of test antigen he might see the same kind of reactions that Tremaine saw in the corneas. I might add also that the small numbers of cells used by Rosenberg et al. apparently cannot transfer delayed hypersensitivity. At least all instances described make use of the cells pooled from several donors to sensitize one recipient, whereas Rosenberg used cells from one recipient to sensitize a number of donor sites.

So far I have only replied to your first paragraph. Your next one takes up the question as to whether perhaps only a single molecular kind of antibody is produced no matter what the inciting conditions (at least that is how I interpret your statement), but that this antibody must be closely regulated in amount and timing of release if it is to be totally absorbed by the tissues leaving no detectable amounts in the serum. I don't think that this is so. If one uses, let us say, a protein with lipopolysaccharide good antibody induction as well as delayed hypersensitivity apparently to the same antigen is produced almost simultaneously. Of course, it is true that these proteins are not single molecular species and one could say that the delayed response is directed against perhaps a minor component of the tuberculoprotein whereas the antibodies are directed against some other component or components, but this doesn't seem likely.

You next take up the matter of Brent's work. Our paper is not intended to criticize the viewpoint that the homograft immunity may be a form of delayed hypersensitivity. Brent may well be right; a recent paper by him and others in the Lancet, September 13, 1958, page 561, presents pretty good evidence that perhaps the reactivity

is of this type in guinea pigs. These kinds of observations, of course, fail to account for the observed fact that agammaglobulinemic patients, who are virtually unable to produce antibodies, but can develop delayed hypersensitivity, still are able to accept homografts, a fact which would make it appear that antibodies are important in the anti-homograft reaction, at least in the human being. I suppose that the solution to this problem has not yet been found.

I believe that it is perfectly feasible that the  $H_2$  genes may reflect different antigenic substances and that tissues other than erythrocytes may possess an antigen or antigens which the erythrocytes don't have. There are many examples of this; a well-known one is the case of the Forssman antigen in guinea pigs. This occurs in all tissues except the red blood cells. But I think that your idea that Brent should try injections of erythrocytes along with a heterologous tissue in order to find whether a T-provoking "principle" might be present in such tissue is a good one. He has tried red cells along with lipopolysaccharide and tubercle bacilli; he was up to see me a couple of years ago while working with Owen at Cal Tech, and obtained material for this purpose. The results were negative.

I believe that Brent and his co-workers have tried to "unify" their work with other work in immunology and allergy. The trouble is that unification has so far been very difficult, for others in addition to Brent. For example, many people have had a good deal of difficulty in the past fifty years in trying to determine definitively whether or not a cellular immunity exists to the tubercle bacillus, in contradistinction to the antibodies which are of course found during the course of the infection. The problem is an analogous one, and people steeped in conventional immunology have so far failed to answer it satisfactorily.

I too look forward to being able to discuss these questions with you firsthand. Meanwhile, if you have

Dr. Lederberg

4

November 11, 1958

the time and inclination, I should like very much to see whether we can communicate effectively on paper.

Best regards to you and your wife.

Sincerely yours,



Sidney Raffel

SR:RB

P.S. I just received an extra copy of pageproof; you needn't bother about the manuscript.

*P.S. - Sorry about the inordinate  
length of this letter. S.*